

Bacteria Monitoring in Surface Waters - Methods

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Introduction

Bacteria monitoring can be an important element of a volunteer monitoring program, providing essential information about human health risks associated with drinking and recreational water contact. In fact, learning whether a particular waterbody is “safe” is often what encourages citizens to get involved with monitoring. Ensuring that the field procedures and laboratory methods used are appropriate for your data requirements and use is key to program success, and the focus of this factsheet.

This factsheet provides a summary of how fecal indicator bacteria are monitored, from collecting samples through laboratory analyses. It includes examples of protocols used by Extension and other volunteer monitoring programs. Links to many helpful resources are highlighted, but several were particularly useful and form the basis for much of this module, making them your first stop for additional details. They are: Building Capacity of *E. coli* Monitoring by Volunteers: A Multi-State Effort (<http://www.usawaterquality.org/volunteer/EColi/index.html>), California Source Water Ambient Monitoring Program – Clean Water Team Citizen Monitoring Program – Compendium for Watershed Monitoring and Assessment (3.4. Bacteria (Pathogen Indicators) http://www.swrcb.ca.gov/water_issues/programs/swamp/cwt_guidance.shtml and the USGS *Field Manual–Fecal indicator bacteria*: <http://water.usgs.gov/owq/FieldManual/Chapter7/7.1.html>.

Bacterial Monitoring Study Design

Determining where to monitor bacteria should be part of your overall monitoring study design (see <http://www.volunteermonitoring.org/GuideBook/DesigningYourStrategyIV.pdf> and <http://water.epa.gov/type/rsl/monitoring/vms22.cfm> to learn more). Since bacteria is typically monitored to determine whether a site is considered safe for swimming, and bacteria levels can vary significantly within a waterbody, bacterial monitoring sites often target swimming beaches, and may differ from sites selected to assess overall water quality. Since a sampler typically can't be used (because they aren't sterile), sites should have safe access to the water so volunteers can use the “one arm” (their arm) sampler. Alternatively pole samplers can be used. These involve attaching a sterile bottle to a pole using a variety of methods as such tape, hook and loop fasteners, rubber bands or other holders. Streamkeepers of Clallam County have created an extendable, adjustable grab-sampling pole that is quite adaptable to a range of uses. (figure 1 , materials list and assembly instructions can be found at www.clallam.net/streamkeepers/assets/applets/poleInstructions.pdf) Careful consideration of sample transportation to the laboratory needs to be done during the study design process and may affect site selection since the maximum sample hold time before analysis begins is typically 6 to 24 hours.

Two excellent resources to help you design your bacteria monitoring program are:

Bacteria Monitoring Protocol Muskoka Watershed Council - http://www.muskokawatershed.org/wp-content/uploads/2011/12/MWC_Bacteria_Protocol1.pdf

Citizen's Guide to Bacteria Monitoring in Vermont Waters - http://www.vtwaterquality.org/lakes/docs/lp_citbactmonguide.pdf.

Figure 1. Pole Sampler



Bacteria Monitoring - Methods

Sampling for Bacteria

Since bacteria are everywhere, great care must be taken to avoid contamination when collecting water samples for analysis. Water sample containers must be sterile and non-toxic. Plastic bottles that have been autoclaved to sterilize them are most frequently used. Sterile plastic bags (e.g. Whirl-pak) or disposable specimen cups are often used by program without access to an autoclave (an autoclave is like a giant pressure cooker where objects are sterilized inside a chamber at high temperature and pressure). Because water sampling devices usually can't be sterilized, samples are usually collected directly into the sterile sampling container. As with all water monitoring, volunteers should practice good sample handling procedures, preferably washing their hands prior to monitoring, and wearing latex or nitrile gloves if they expect to sample in a contaminated area (see www.volunteermonitoring.org/E_Coli_Bacteria_Fact_Sheet.pdf for resources on good hygiene practices).

Open the container just before sampling, being very careful not to touch the inside of the container or the lid with anything other than the water sample itself. Close the sample container either underwater or immediately after collection. Store the samples on ice or with cold packs during transportation and until delivery to minimize bacterial population growth after collection. In the lab, samples should be refrigerated or kept on ice until analysis, which should be within six to twenty-four hours of collection. (See <http://water.epa.gov/type/rsl/monitoring/vms511.cfm> for a more detailed overview.)

Field Sampling Protocol Guidance:

Citizens Monitoring Bacteria: A training manual for monitoring *E. coli* (Chapters 3 & 4): http://www.volunteermonitoring.org/Final_ecoli_06may06.pdf

Maine Healthy (Coastal) Beaches Program: Training Plan for Collecting Water Samples and Recording Environmental Data: <http://www.mainehealthybeaches.org/documents/MHB%20Training%20Plan08.pdf>

Surface Water Ambient Monitoring Program Clean Water Team: http://www.swrcb.ca.gov/water_issues/programs/swamp/docs/cwt/guidance/3410.pdf (slides 38 - 59)

Surfrider Foundation - Blue Water Task Force Coastal Water Quality Monitoring Manual http://public.surfrider.org/BWTF_manual_June2003.pdf



Other Guide to Growing Programs - Bacteria Modules

See ***Volunteer Monitoring of Bacteria*** to learn more about:

- What are Bacteria
- Why Monitor Fecal Indicator Bacteria
- Which Bacteria Should You Monitor
- Emerging Indicators

See ***Presenting Bacteria Data Effectively*** to learn more about:

- Unique Characteristics of Bacteria Data
- Effective Charts and Graphs for Presenting Bacteria Data
- Cautions Regarding the Use of Bacteria Data

Photo credits:

Fig. 1 Clallam County Streamkeepers

Fig. 2 Murulee Byappanahalli, USGS

Fig. 3 Roland Park County School

Fig. 4 edusanjalmicro.blogspot.com

Fig. 5 Skyline College - Bio 675

Fig. 6 Laboratorios Condo

Fig. 7 Weber Scientific

Fig. 8 Micrology Laboratories

Fig. 9 Hach

Fig. 10 Avensys Solutions

Fig. 11 USDA

Fig. 12 Florida Atlantic University

Fig. 13 USGS

Fig. 14 US EPA

Fig. 15 Wisconsin Extension

Fig. 16 Georgia Adopt-A-Stream

Bacteria Testing Laboratory Basics

Determining whether to use a laboratory (either a commercial lab or perhaps one at a local sewage treatment plant) or to set-up your own will largely depend your data objectives and program resources. Consulting with potential data users is critical as the use of specific procedures or of a certified laboratory may be required in order for state agencies and others to use your data. (See <http://www.volunteermonitoring.org/pdf/GuideBook/PresentingBacteriaDataXV.pdf> for details on ensuring that your data is reported properly.)

Having your own laboratory can provide greater flexibility in the frequency and timing of sample collection since you won't have to coordinate with another lab's hours of operation. It also makes follow-up tests easier, and can offer opportunities for volunteers that might be interested in indoor activities to participate in your program. Of course maintaining your own lab does require an initial investment in specialized equipment. Most methods require incubation at temperatures of 35 to 44.5 °C, which means the use of incubators or waterbaths. Some methods require very precise temperatures, which increases the sophistication and thus the cost, of the incubator. These equipment and procedures mean additional lab space and training for operators, and time to ensure that tests are being run according to your standard operating procedures *and* that the appropriate quality assurance and quality control (QA/QC) measures are used. This all has to be factored into the budget along with the purchase of media and other consumables needed for the analyses.

Working with commercial labs negates the need for the equipment, space and operator, but typically cost more per sample and may have limited hours during which you can drop off samples for analysis. If you are able to work with a municipal lab that provides free analyses (such as a sewage treatment plant) then you may also be limited in the number of, and timing of samples you can have processed. If you choose to work with a commercial or municipal lab, it is important that you know the analytical and QA/QC methods they use, as well as detection limits, both minimum and maximum values. You should also work with them to ensure you follow all of their chain of custody requirements.

In the laboratory, samples are most commonly processed by membrane filtration, multiple tube fermentation or chromogenic/fluorogenic substrate methods. Some methods use combinations of these to produce results. While these methods all reproduce aspects of the environment the bacteria are commonly found in (i.e., the intestines of warm blooded animals), they vary significantly in the equipment and processing/incubation time required in order to test for specific species (see Southern California Coastal Water Research Project (<http://www.sccwrp.org/researchareas/BeachWaterQuality.aspx> or the Hach Bacteria Test Guidelines for a synopsis of methods and basic guidelines for proper lab techniques – <http://www.hach.com/asset-get.download-en.jsa?id=7639984414>). If you are interested in learning about other methods see the National Environmental Methods Index (NEMI) <http://www.nemi.gov>, “A comparison of ten USEPA approved total coliform/*E. coli* tests (<http://www.iwaponline.com/jwh/005/0267/0050267.pdf>) or USEPA Bioindicators – Clean Water Act Analytical Methods at <http://www2.epa.gov/cwa-methods/other-clean-water-act-test-methods-microbiological>.

Incubator Options

Laboratory incubators cost from \$300 to \$3500, with the more precise units (constant temperature +/- 0.2 °C) generally costing at least \$1000. If the method that you have selected does not require the use of a laboratory incubator (allows for at least +/- 2 – 5 °C temperature fluctuation during incubation), there are several low-cost alternatives.

- Styrofoam-box-type egg hatching incubators such as Little Giant and Hova-bator (available through farming and biological science suppliers).
- Home-made incubator using a cardboard box or portable cooler with a 40-watt to 100-watt light bulb (depending on container size and temperature needed – you'll need to experiment). The bulb can be on a small reading light or work light placed in the box. A thermometer should be used to monitor temperature so you can regulate it. Vents can be cut to allow for regulation if using a cardboard box or Styrofoam cooler.

Figure 2. Membrane filtration



The membrane filtration (MF) method is a well-established method approved by most federal and state agencies to assess bacterial concentrations (see <http://water.usgs.gov/owq/FieldManual/Chapter7-Archive/Chapter7.1/7.1.3.html> for details). Water is pulled through a filter that traps all the bacteria from the sample (figure 2). The filter is then placed in a petri dish with growth medium and incubated at a specific temperature (usually 35, 37, 41 or 44.5 °C depending on the media being used) which is approximately the temperature of the intestines of warm-blooded animals (where these organisms normally grow). The resultant bacterial colonies that grow are visible to the human eye and easily counted. Varying the type of growth medium, temperature and incubation periods

help laboratories to isolate particular species of bacteria. Results are reported as the number of colony forming units (cfu) per volume of sample because only viable, or bacteria capable of reproducing, cells are counted.

Samples exposed to ultra-violet light (including sunlight) or to other stressors may produce values lower than the actual fecal indicator bacteria concentration. In addition, because you must be able to discriminate between colonies, samples that are very turbid or have very high concentrations of bacteria can be difficult to analyze since colonies grow together or silt particles can make it impossible to clearly identify colony edges. An ideal counting range is 20 – 80 colonies. If the anticipated bacterial count is unknown or expected to be higher than 80, multiple plates, possibly even serial dilutions, must be analyzed for each sample in order to ensure a valid result. For example, if your sample is very silty you may decide to filter only 1 mL of sample. If only 1 colony forms, we would report that as 100 mL bacteria /100 mL of water ((Count / vol filtered)* 100). However with that sample volume, the minimum level at which bacterial concentration could be determined would be 100. The smaller the sample volume used, the higher the minimum level of detection, often higher than the criteria used to assess whether water is safe for contact. A high minimum limit of detection for very silty or contaminated samples can limit the value of MF methods under those conditions.

The pour plate method is similar to membrane filtration since the end point is counting of visible colonies. However with pour plates, media is added to a volume of sample, mixed well and poured into a petri plate (figure 3). The agar in the media gels, and the plate is incubated at a designated temperature for 24 – 72 hours. (figure 4). The media and temperature used are selective for specific types of bacteria. The colonies counted may permit further selection by only counting various colors of colonies or only those with gas bubbles, etc. depending on the media used. Petrifilm uses a modified version of the pour plate method.

Figure 3. Pouring a plate

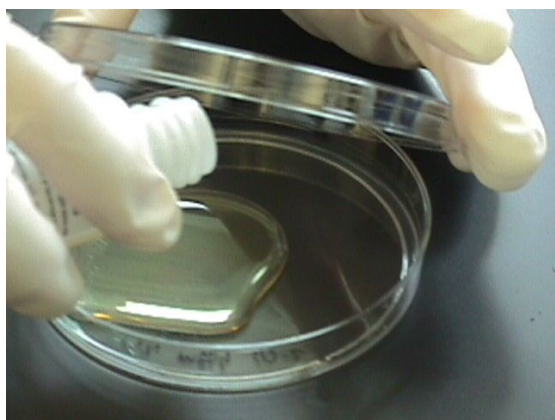
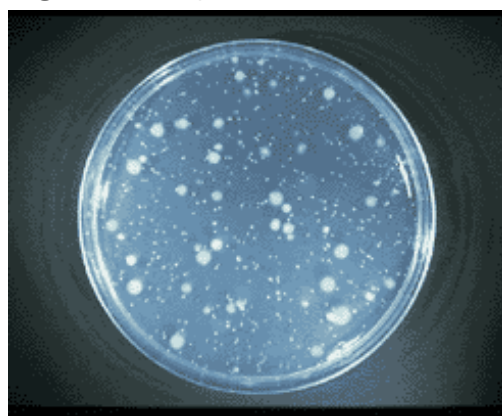


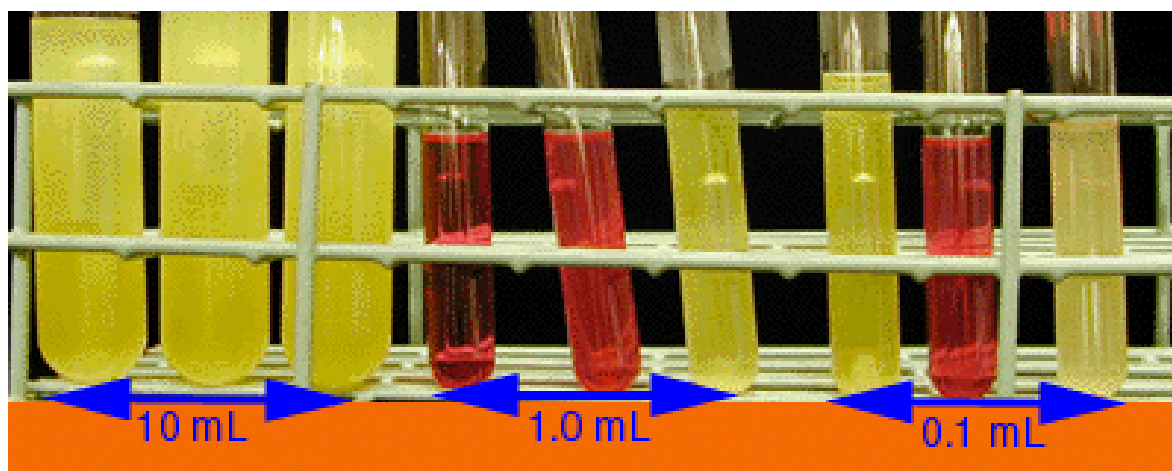
Figure 4. Pour plate after incubation



Laboratory Methods Overview (continued)

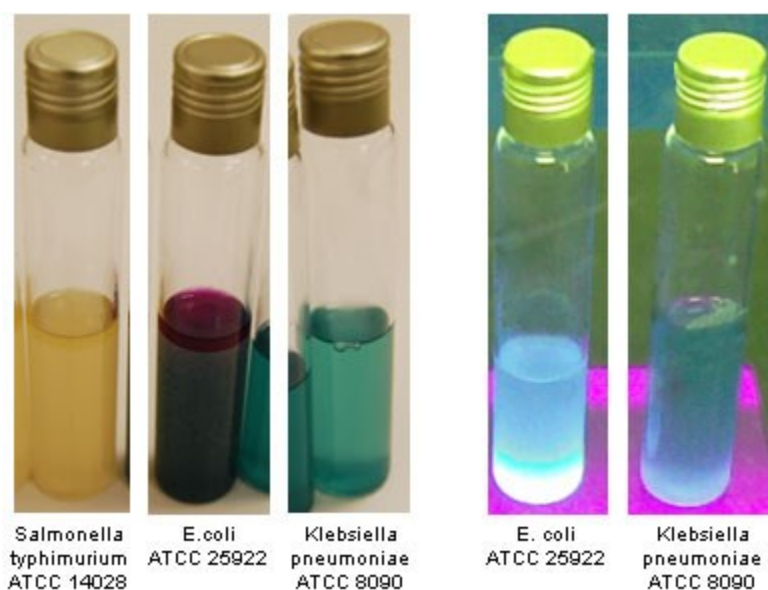
The multiple tube fermentation (MTF) method relies on the fact that coliform bacteria produce gas when fermenting, or breaking down sugars. With MTF a series of tubes with media that select for specific bacteria are inoculated with various volumes of sample water. After a specified incubation time (usually 24 to 48 hours) at a defined temperature, the tubes are inspected for the presence of gas (figure 5). An estimate of the bacterial density is determined from the number of positive (gas producing) tubes in each series. The MTF method is very labor and glassware intensive, and is being rapidly replaced by other methods. And unlike MF methods, MTF methods don't provide a direct count of bacteria. Rather, they are based on a statistical probability that the sample contained a certain number of bacteria based on the number of tubes at particular sample volumes showing a presence of the indicator bacteria. Referred to as the Most Probable Number (MPN) the results are compared to a statistically-derived MPN table, and reported as the number of organisms per 100 ml of sample. (See <http://www.jlindquist.net/generalmicro/102dil3.html> for details on the MPN process and an MPN table. See <http://www.unc.edu/depts/case/BMElab/MPNcalculator/MPNequations.doc> for the equations behind the MPN tables.)

Figure 5. Multiple Tube Fermentation



Chromogenic and fluorogenic substrate (CFS) methods also rely on the metabolic differences between bacteria. With these techniques, enzymes found in various species react with substrates to either change the color (chromogenic) of the media or colonies, and / or to make the media or colonies fluoresce (fluorogenic) when exposed to a specific wave length of ultra-violet (UV) light. Some enzyme-based methods, such as those that use defined substrate technology, also inhibit non-coliform bacteria growth, and thus non-coliform bacteria can't interfere with the recovery of coliforms. This technology is based on the principle that typically *only* the target microbe can utilize vital nutrients from the media. The CFS methods, which are usually commercially available patented kits or media, can be either presence/absence or quantification tests, and may be approved only for limited uses.

Figure 6. Chromogenic and Fluorogenic Substrates



Bacteria Monitoring - Methods

Table 1. Comparison of Established Cultivation Methods (Adapted from Köster et al.)

Technique	Advantages	Disadvantages
Presence/ absence (P/A) test using liquid chromogenic or fluorogenic media (e.g. Colisure)	<ul style="list-style-type: none"> • Flexible sample volume range, • Applicable to all kinds of samples, • Allows resuscitation and growth of injured organisms, • Usually easy interpretation of test results and no special skills required, • Minimal time and effort needed to start the test, • Precision and sensitivity can be chosen by selection of volumes analyzed, multiple dilution levels and/or replicates, • Media often inexpensive. 	<ul style="list-style-type: none"> • In routine application, when few replicates are used, the precision is often low, • When the selectivity of the medium is not adequate, the target organisms can be masked due to the growth of other microorganisms, • Sample may contain inhibitors affecting the growth of the target organisms, • For the isolation of pure cultures, further cultivation on solid media is necessary, • No information on level of concentration of target organisms.
Most probable number (MPN) (e.g. modified IDEXX Quanti-tray)	<ul style="list-style-type: none"> • Same as for P/A test above • Information on the level of concentration of target organisms is obtained. 	<ul style="list-style-type: none"> • Same as for P/A test above • Media can be expensive, • If multiple sample volumes are run, media and other supply costs, as well as incubation space needed, increases.
Pour plate/direct inoculation (e.g. Coliscan Easygel or Petrifilm)	<ul style="list-style-type: none"> • Simple and inexpensive method, • Equipment needs generally low. 	<ul style="list-style-type: none"> • Maximum 1 ml sample volume routinely analyzed yields low resolution and high limit of detection, • Interpreting colonies not easy
Membrane filtration (e.g. mTEC or Coliscan MF)	<ul style="list-style-type: none"> • Flexible sample volume range enabling the use of large sample volume and therefore increased sensitivity • Water soluble target organism growth inhibitors separated from the sample in the filtration step • Quantitative result and good precision if the number of colonies grown adequate 	<ul style="list-style-type: none"> • Quality of membranes varies, • Solid particles and chemicals adsorbed from sample to the membrane during filtration may interfere with the growth of the target organism, • Very high detection level or not valid with turbid samples, • Interpreting colonies not always easy.

Specific Methods

Coliscan (Micrology Laboratories - <http://www.micrologylabs.com/page/50/Our-Methods> LaMotte Company - www.lamotte.com/en/microbiological/coliform) media test total coliforms and *E. coli* specifically with formulations for use in two distinct ways. A pour plate method for use with surface water (i.e., lake, river, etc.) samples mixes 1 – 5 ml of water directly into the media, which is poured into a petri dish and incubated at various temperatures (figure 7). For water with low expected bacteria levels (such as drinking water) larger sample volumes (up to 100 ml) are filtered through a membrane filter, which is placed on a pad saturated with Coliscan MF liquid media

Figure 7. Coliscan Easygel plate



in a petri dish (figure 8). The colonies that grow either within the Easygel or on the surface of the filter are counted. With both formulations, total coliform bacteria will grow as pink-magenta colonies while *E. coli* will grow as purple-blue colonies. Other bacterial types will generally grow as non-colored or pink-tan colonies. However, differentiating between these colors can be a challenge, particularly for volunteers with color-blindness. These methods are unique in that the plates will produce results if the plates are incubated at room temperature for up to 48 hours, and therefore don't require the use of expensive laboratory incubators. But the results may not be as comparable with reference laboratory results and the volunteers reported that it was difficult to interpret the plate (O'Brien, 2006). Using a 35 °C incubator allows the test to be read in 24 hours, and produces more comparable results, thus if your data objectives requires you to meet USEPA approved requirements, you'll need to use a 35 °C incubator. Values are reported as CFUs. (See Montana's Blue Water Task document http://www.bluewatertaskforce.org/documents/BWTFCWQMP_SOP_FINAL_284.pdf for an example of standard operating procedure for this method.)

Figure 8. Coliscan MF filter

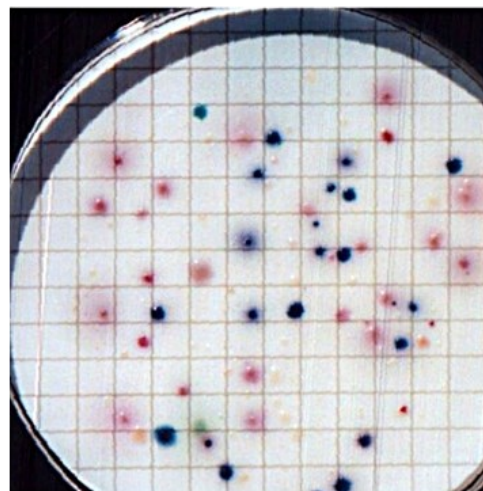


Figure 9. m-ColiBlue24 plate



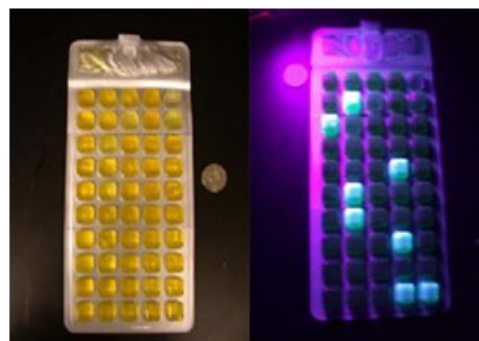
m-ColiBlue24 (Hach - <http://www.hach.com/m-colibblue24-broth-glass-ampoules-pk-20/product?id=7640249625> and Millipore -http://www.merckmillipore.com/INTL/en/product/m-ColiBlue24@-Broth,MM_NF-MOOPMCB24) is prepared broth for use with membrane filtration to test for total coliforms and *E. coli*. It is very similar to Coliscan MF. It can be used with sample volumes of up to 100 ml and is incubated at 35 °C for 24 hours. Coliform colonies are red, and *E. coli* colonies are blue, with the total coliform count being the sum of those reported as CFUs (figure 9).

Colisure / Colilert / Colilert-18 IDEXX (<http://www.idexx.com/water/water-testing-solutions.html>) are chromogenic and fluorogenic substrate methods. These reagents can be used either as presence / absence (P/A) tests, or as quantification tests. In both cases, the reagent is added to a sample volume of up to 100 ml. When performing P/A tests, the bottles are incubated at 35 °C for 24 hours after mixing (Colisure can be incubated for 24 - 48 hours); if the media/sample mixture is yellow after incubation, the sample is negative for total coliforms and *E. coli*. If it's red/magenta the sample is positive for total coliforms (figure 10). To determine whether *E. coli* are present, the samples which test positive for total coliforms (red/magenta) are viewed within 5 inches of a *365 nm UV light in a dark environment, if they fluoresce, they are positive for *E. coli*. Colilert and Colilert-18 are very similar except that the end points are: yellow is positive for total coliform, and with fluorescence it's *E. coli* (figure 11). The total incubation and time during which the samples can be read is less for these two methods. It is only 24 - 28 hours with Colilert and 18 - 22 hours with Colilert-18.

Figure 10. Colisure



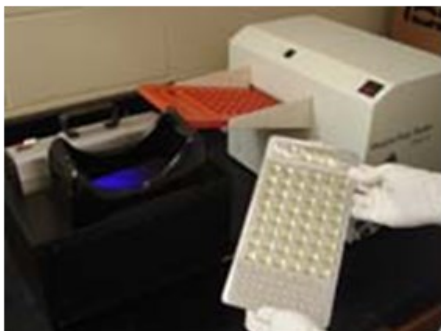
Figure 11. Colilert in a Quanti-Tray



*Note: You should never look directly into UV lamps. Eye damage can occur if they are exposed to direct ultraviolet radiation.

These media can be used to quantify total coliform and *E. coli* through the use of a proprietary method based on MTF. The IDEXX Quanti-Tray method substitutes a plastic tray and sealer for the tubes (figure 12), and an IDEXX-generated MPN table for use with the trays. To complete this test, the mixed sample and appropriate reagent are poured into a Quanti-Tray, or Quanti-Tray/2000 (if bacteria counts are expected to exceed 200 per 100 ml). The tray is placed in a rubber insert, which is then put through a heated sealing unit. The sealed trays are incubated at 35 °C. Positives are red/magenta or yellow for total coliform with fluorescence when viewed under a 365 nm UV light indicating *E. coli*. The number of positive cells is then compared to the IDEXX MPN table (http://www.idexx.com/pubwebresources/pdf/en_us/water/qt51mpntable.pdf or http://www.idexx.com/pubwebresources/pdf/en_us/water/qt97mpntable.pdf).

Figure 12. Quanti-Tray Sealer

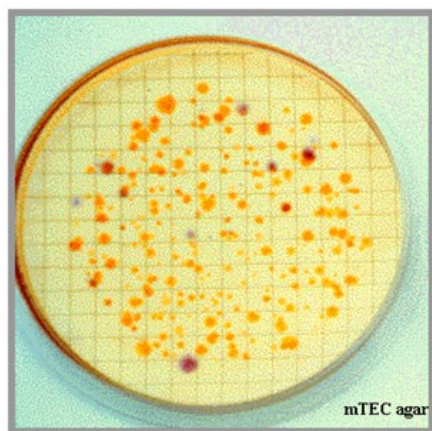


Enterolert, another IDEXX product, detects and quantifies enterococci with the use of the Quanti-Tray system. The process is the same as with the Coli-products, but the trays are incubated at 41 °C, with an incubation time of 24-28 hours. After incubation, the trays are read under the UV light with the fluorescing cells counted as positive for enterococci, and the counts compared to the MPN table. The IDEXX methods are accepted for use by many state and federal agencies, are very easy to use and allow volunteers to produce results that are comparable to those of certified laboratories. In fact the *E. coli* Project found that volunteers preferred the IDEXX methods and produced very good results (<http://www.usawaterquality.org/volunteer/EColi/ResultsProducts.htm>). The drawback with these methods was the cost. The

reagent and trays average more than \$5 per sample, with the initial investment for the sealer and incubators in excess of \$4000.

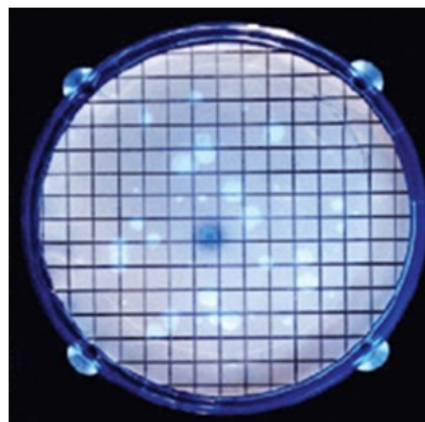
Membrane filtration onto mTEC media is sometimes considered as the “gold standard” for *E. coli* testing as it was the method used in the USEPA epidemiological studies that helped establish the 1986 *E. coli* and enterococci criteria. It is commonly used in method validation studies. The mTEC media, and a newer version called “modified mTEC” are selective for fecal coliform bacteria, which is enhanced through the use of an incubation temperature of 44.5 °C to kill off non-fecal organisms. This method is more equipment and labor intensive because it requires a 2-hour “recuperative” incubation at 35 °C, followed by 22 hours at precisely (+/- 0.2 °C) 44.5 °C. After incubation, using the original mTEC, the filter is placed on a pad saturated with urea and the number of yellowish colonies are counted. The urea step is skipped with the modified mTEC, and the red/magenta colonies are counted. The extra steps and precise (i.e., expensive) incubator required makes these methods less convenient for volunteer programs. However, USEPA has created MI media, which is incubated at 35 °C (+/- 0.5) for 20 - 24 hours. The filter is placed under a UV lamp, with the fluorescing colonies counted as total coliforms. Colonies that fluoresce and are blue, are counted as *E. coli*. The need for only one incubator set at 35 °C (+/- 0.5) reduces some cost and time barriers to its use by volunteer programs.

Figure 13. Filter on mTEC media



- *E. coli* colonies remain yellow after the urease test
- non-*E. coli* colonies turn from yellow to purple from the breakdown of urea

Figure 14. Filter on MI media



Specific Methods (continued)

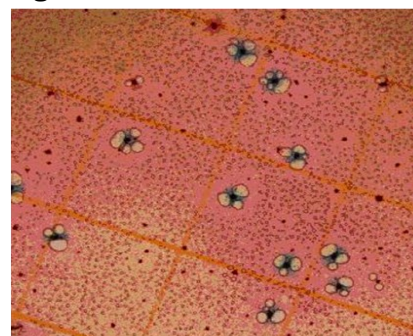
Petrifilm is a unique commercial product from 3M Microbiology for counting *E. coli* (3M has film media for other bacteria species available) through a direct plating method. Petrifilm is a circle of prepared media on a gridded paper with a translucent film cover. To inoculate, the top film is lifted and 1 ml of sample or diluted sample is placed in the center of the media using a sterile pipette (figure 11). The top film is then carefully rolled onto the sample to prevent trapping any air bubbles, and smoothed out with a spreader. The “plates” are incubated in stacks of up to 20 in an incubator for 24 hours at 35 °C. After incubation, blue colonies with gas bubbles are counted as *E. coli* (figure 16). Because of the low sample volume used, multiple plates per sample should be analyzed if relatively low counts (<1000/100 ml) are anticipated. Petrifilm is typically the least expensive of methods commonly used by volunteers, facilitating replicate plates (<http://.usawaterquality.org/volunteer/EColi/KitCosts.htm>). It is important to note that Petrifilm is not an EPA-approved method for water testing. However it performed well in a comparison study and thus could be a useful tool for bacterial screening in surface waters (O'Brien,2006).

Regardless of whether you choose to work with a commercial lab or analyze the samples in-house, the bacteria and method you choose should meet your data objectives. In addition, the appropriate QA/QC steps should be used. See our guidance module “Building Credibility: Quality Assurance and Quality Control for Volunteer Monitoring at <http://www.volunteermonitoring.org/GuideBook/BuildingCredibilityVI.pdf> to learn more.

Figure 15. Inoculating Petrifilm



Figure 16. *E. coli* colonies in Petrifilm



Comparison of Methods:

Volunteers Conduct Bacteria Methods Comparison Study (http://water.epa.gov/type/rsl/monitoring/upload/2006_03_20_monitoring_volunteer_newsletter_volmon18no1.pdf)

Comparison of Beach Bacterial Water Quality Indicator Measurement Methods (<http://lshs.tamu.edu/docs/lshs/end-notes/BeachBactWaterqQual-0599552031/BeachBactWaterqQual.pdf>)

Comparison among IDEXX, Membrane Filtration and Multiple Tube Fermentation Bacterial Indicator Analysis Methods (<http://www.sccwrp.org/ResearchAreas/BeachWaterQuality/ComparisonAmongIDEXMembraneFiltration.aspx>)

Comparing *E. coli* Results Analyzed by Colilert® and Membrane Filtration (<http://www.orsanco.org/images/stories/files/programs/WatershedPollutantReduction/docs/colilertvsmembranefiltrationcomparison.ppt>)

Comparison of bacterial indicator analysis methods in stormwater-affected coastal waters (<http://www.vliz.be/imisdocs/publications/55411.pdf>)

Summary

Because understanding the source of contamination is so important in determining the most effective means of reducing or eliminating fecal pollution, being able to quantify sources has become the goal of many researchers. This factsheet detailed pros and cons of a number of methods commonly used by volunteer monitoring programs for assessing surface water. Currently available methods range from inexpensive, easy-to-use screening tools, to very precise, more expensive methods that are acceptable to numerous agencies, and most points in between. Selecting the method that best meets your data quality objectives and uses, as well as your technical, financial and other resources, is critical for program success.

Bacteria Monitoring - Methods

References and for further information on bacterial monitoring:

- California Source Water Ambient Monitoring Program – Clean Water Team Citizen Monitoring Program – Compendium for Watershed Monitoring and Assessment (3.4. Bacteria Pathogen Indicators http://www.swrcb.ca.gov/water_issues/programs/swamp/cwt_guidance.shtml)
- E. coli Project. Building Capacity of E. coli Monitoring By Volunteers: Methods Manual <http://www.volunteermonitoring.org/Ecoli/>
- Ely, E. 1998. Bacteria Testing. *The Volunteer Monitor*. Volume 10, No. 2. http://water.epa.gov/type/rsl/monitoring/upload/2004_10_13_monitoring_volunteer_newsletter_volmon10no2.pdf
- Ely, E. Editor. 2006. *The Volunteer Monitor* – Bacteria Monitoring, Volume 18, Number 1, Winter 2006; (http://water.epa.gov/type/rsl/monitoring/upload/2006_03_20_monitoring_volunteer_newsletter_volmon18no1.pdf).
- Heufelder, G. 1997. Interpreting Fecal Coliform Data: Tracking Down the Right Sources. *The Volunteer Monitor*. Volume 9, No. 2. http://water.epa.gov/type/rsl/monitoring/upload/2004_10_13_monitoring_volunteer_newsletter_volmon09no2.pdf
- Köster et al. 2003, 'Analytical methods for microbiological water quality testing', in *Assessing Microbial Safety of Drinking Water: Improving Approaches and Methods*, OECD/WHO Drinking Water Quality Series, IWA Publishing, London, pp. 237-295. Chapter 8 Analytical Methods for Microbiological Water Testing, http://www.who.int/water_sanitation_health/dwq/en/9241546301_chap8.pdf
- Miceli, G.A. 1998. Bacterial Testing Q & A. *The Volunteer Monitor*. Volume 10, No. 2. (see above)
- Scott, T, J. Rose, T. Jenkins, S. Farrah and J. Lukasik. 2002. Microbial Source Tracking: Current Methodology and Future Directions. *Applied and Environmental Microbiology*. December 2002. Vol. 68, No. 12, pp. 5796-5803 (<http://aem.asm.org/content/68/12/5796>)
- Ohrel Jr., R.L. and K.M. Register. 2001. *Volunteer Estuary Monitoring: A Methods Manual*. Second Edition. U.S. EPA. http://water.epa.gov/type/oceb/nep/upload/2007_04_09_estuaries_monitoruments_manual.pdf
- Pathogen Indicators of Recreational Water Quality (a PowerPoint Presentation) <http://www.state.nj.us/dep/wms/Recreational%20Indicators%20Overview.pdf>
- O'Brien, E. 2006. Volunteers Conduct Bacteria Methods Comparison Study. *The Volunteer Monitor* – Volume 18, Number 1, Winter 2006; (http://water.epa.gov/type/rsl/monitoring/upload/2006_03_20_monitoring_volunteer_newsletter_volmon18no1.pdf).
- Turin, D. and M. Liebman. 2002. Keeping Posted: Communicating Health Risks at Public Beaches. *Journal of Urban Technology* 9:45-69.
- USEPA. 1986. The Ambient Water Quality Criteria for *Bacteria* – 1986. EPA440/5-84-002 January, 1986
- USEPA. Beaches Environmental Assessment and Coastal Health (BEACH) Program http://water.epa.gov/type/oceb/beaches/beaches_index.cfm.
- USEPA. Water: Monitoring & Assessment 5.11 Fecal Bacteria <http://water.epa.gov/type/rsl/monitoring/vms511.cfm>
- USGS Field Manual – Fecal indicator bacteria: <http://water.usgs.gov/owq/FieldManual/Chapter7/7.1.html>

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